CLAIMS

What is claimed is:

1. A process for isolation of epothilone B from an epothilone-producing microorganism comprising:

(a) fermenting a strain of epothilone-producing microorganism in the presence of a resin that adsorbs epothilone B by hydrophobic interaction;

(b) collecting the resin in a water-based medium;

(c) extracting the resin with a solvent selected to extract epothilone B and to separate it from the water-based medium; and

(d) crystallizing epothilone B from the extraction phase prior to a chromatography step.

2. The process of claim 1 wherein the crystallized epothilone B from step (d) is substantially pure.

3. The process of claim 1 wherein the resin is extracted with a polar solvent.

4. The process of claim 1 wherein said fermentation step further comprises feeding an additive capable of improving the amount of epothilone B produced as compared with the amount of epothilone A produced.

5. The process of claim 4 wherein the additive is TASTONE™, maltrin or glycerol.

6. The process of claim 1 wherein said fermentation step comprises continuously feeding an additive capable of improving the ratio of epothilone B to epothilone A.

7. The process of claim 4 wherein said additive is a propionic acid salt or ester.

8. The process of claim 7 wherein said additive is sodium propionate, propionic acid methyl ester or propionic acid ethyl ester.

9. The process of claim 1 wherein the crystallization is conducted to reduce the amount of epothilone A to about 55% or less of the amount of epothilone A present after extraction step (c).

10. The process of claim 9 further comprising

(e) at least a second crystallization step effective to reduce the amount of epothilone A to about 55% or less of the amount of epothilone A present after crystallization step (d).

11. The process of claim 1 wherein the epothilone-producing microorganism is *Sorangium cellulosum*.

12. The process of claim 11 wherein said microorganism is *Sorangium cellulosum* strain ATCC No. PTA 3880.

13. The process of claim 11 wherein said microorganism is *Sorangium cellulosum* strain ATCC No. PTA 3881.

14. The process of claim 1 wherein the resin is a styrene/divinylbenzene-based polymer.

15. The process of claim 14 wherein the resin is XAD-16.

16. The process of claim 1 wherein said step (d) comprises:

(i) the addition of a second solvent in which epothilone B is either not soluble or sparingly soluble;

(ii) removing at least a portion of the extraction solvent; and

(iii) transitioning the resultant solvent or solvent mixture to a temperature at which epothilone B crystallizes.

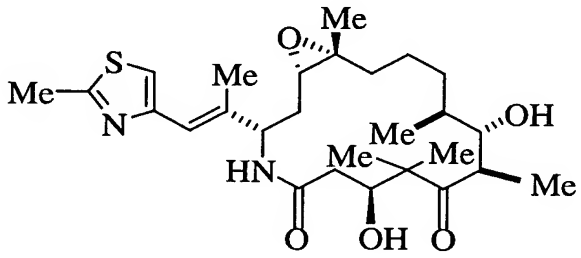
17. The process of claim 16 wherein the extraction solvent is ethyl acetate or MTBE, and the second solvent is toluene.

18. The process of claim 1 further comprising:

(f) prior to step (c), washing the resin with aqueous acetonitrile, or aqueous methanol, or an aqueous medium containing a detergent and an amine reagent added in base form, the aqueous medium selected to not elute epothilone B.

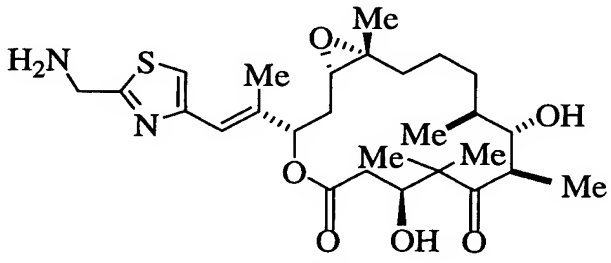
19. The process of claim 1, wherein step (c) further comprises polish filtering the epothilone B containing solvent.

20. The process of claim 1 further comprising converting the epothilone B, or a solvate thereof, to Derivative 2, or a solvate thereof, having the formula:



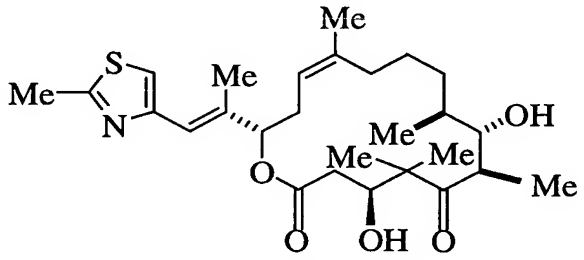
Derivative 2

21. The process of claim 1 further comprising converting epothilone B, or a solvate thereof, to Derivative 1, or a salt or a solvate thereof, having the formula:



D rivative 1

22. The process of claim 1 further comprising converting epothilone B, or a solvate thereof, to Derivative 3, or a solvate thereof, having the formula:



Derivative 3

23. A method for cultivation of a microorganism that produces epothilone A or epothilone B comprising:

feeding a culture of the microorganism being cultivated under conditions selected to promote production of epothilones with propionic acid, a precursor thereof, or a salt of one of the foregoing, wherein the timing of feeding and the amount of propionic acid, a precursor thereof, or a salt of one of the foregoing, are selected to provide at least a two-fold increase in a ratio of epothilone B to epothilone A relative to the ratio of epothilone B to epothilone A produced by a culture of the microorganism cultivated in the absence of feeding of propionic acid, a precursor thereof, or a salt of one of the foregoing; and

isolating epothilone B from the culture.

24. The method of claim 23 wherein the timing of contacting and the amount of propionic acid, a precursor thereof, or a salt of one of the foregoing, are selected to provide at least a three-fold increase in a ratio of epothilone B to epothilone A.

25. The method of claim 23 wherein the timing of contacting and the amount of propionic acid, a precursor thereof, or a salt of one of the foregoing, are selected to provide an increase of the ratio of epothilone B to epothilone A to at least 1.5.

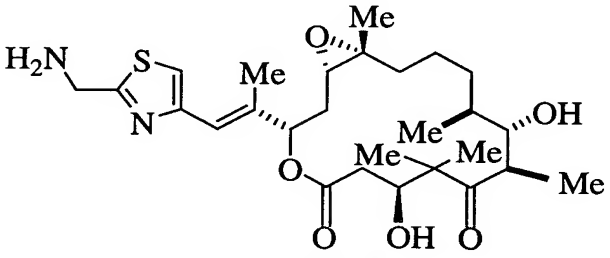
26. The method of claim 23 wherein the microorganism is a strain of *Sorangium cellulosum*.

27. The method of claim 23 wherein the propionic acid, a precursor thereof, or a salt of one of the foregoing, is added during or after the growth phase of the culture.

28. The method of claim 27 further comprising feeding the culture with a vitamin, a mineral, a carbohydrate source or an amino acid source in an amount that increases the amount of epothilone B produced relative to the amount of epothilone B produced in the absence of feeding.

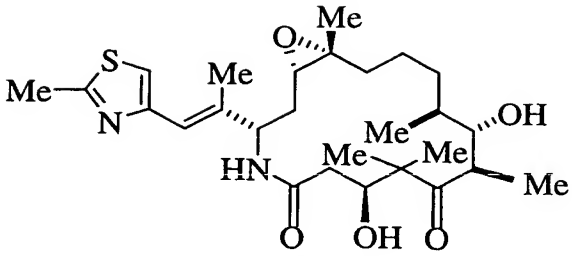
29. The method of claim 27 further comprising feeding the culture with a mixture of monobasic and dibasic phosphate.

30. The method of claim 23 further comprising converting epothilone B, or a solvate thereof, to Derivative 1, or a salt or a solvate thereof, having the formula:



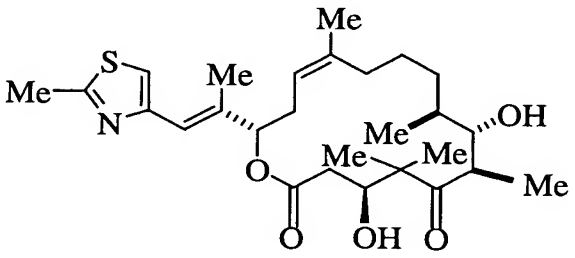
Derivative 1

31. The method of claim 23 further comprising converting epothilone B, or a solvate thereof, to Derivative 2, or a solvate thereof, having the formula:



Derivative 2

32. The method of claim 23 further comprising converting epothilone B, or a solvate thereof, to Derivative 3, or a solvate thereof, having the formula:



Derivative 3

33. A strain of *Sorangium cellulosum* that produces, under epothilone B comparative production conditions, at least 5 mg of epothilone B/g of resin.

34. A strain of claim 33 that produces epothilones with an epothilone B/A ratio of at least 1.0.

35. A strain of *Sorangium cellulosum* deposited as ATCC No. PTA-3880.

36. A strain of *Sorangium cellulosum* deposited as ATCC No. PTA-3881.

37. A method of purifying an epothilone isolated from the method of claim 1 by reverse phase high performance liquid chromatography (HPLC), comprising:

(a) equilibrating a reverse phase HPLC column comprising a separation resin with an aqueous organic solvent or an aqueous mixture of organic solvents;

(b) providing a load sample dissolved in a suitable organic solvent or a mixture of organic solvents;

(c) injecting the column with the load sample, and a trailing volume of a suitable organic solvent or a mixture of organic solvents effective to reduce epothilone precipitation in the loading volume; and

(d) eluting the column with an aqueous organic solvent or an aqueous mixture of organic solvents, that starts with a lower organic content and increases thereafter to more than that of the mixture used in the equilibrating step, to obtain the epothilone.

38. The method of claim 37 wherein the high performance liquid chromatography (HPLC) is performed using an apparatus comprising a loading volume intervening between an injection port and a separation column.

39. The method of claim 37 wherein the organic solvent of step (b) is dimethylsulfoxide.

40. The method of claim 37 wherein the organic solvent of step (c) is dimethylsulfoxide.

41. The method of claim 37 wherein the organic solvent of step (d) is aqueous acetonitrile or aqueous methanol.

42. The method of claim 37 wherein the injecting step comprises using an immediately preceding volume of dimethylsulfoxide effective to reduce epothilone precipitation in the loading volume.

43. The method of claim 37 further comprising:

(e) crystallizing the epothilone to obtain purified epothilone B.

44. A method of purifying an epothilone isolated from the method of claim 1 by normal phase high performance liquid chromatography (HPLC) comprising:

(a) equilibrating a normal phase HPLC column comprising a separation gel or resin with an organic solvent or a mixture of organic solvents;

(b) providing a load sample dissolved in an organic solvent or a mixture of organic solvents;

(c) injecting the column with the load sample; and

(d) eluting the column with an organic solvent or a mixture of organic solvents that starts with a less polar solvent content and increases thereafter to a more polar solvent mixture than that used in the equilibrating step, to obtain the epothilone.

45. The method of claim 44 wherein the organic solvent of step (b) is dichloromethane.

46. The method of claim 44 wherein the organic solvent of step (d) is ethyl acetate or n-heptane.

47. The method of claim 44 further comprising:

(e) crystallizing the epothilone to obtain purified epothilone B.